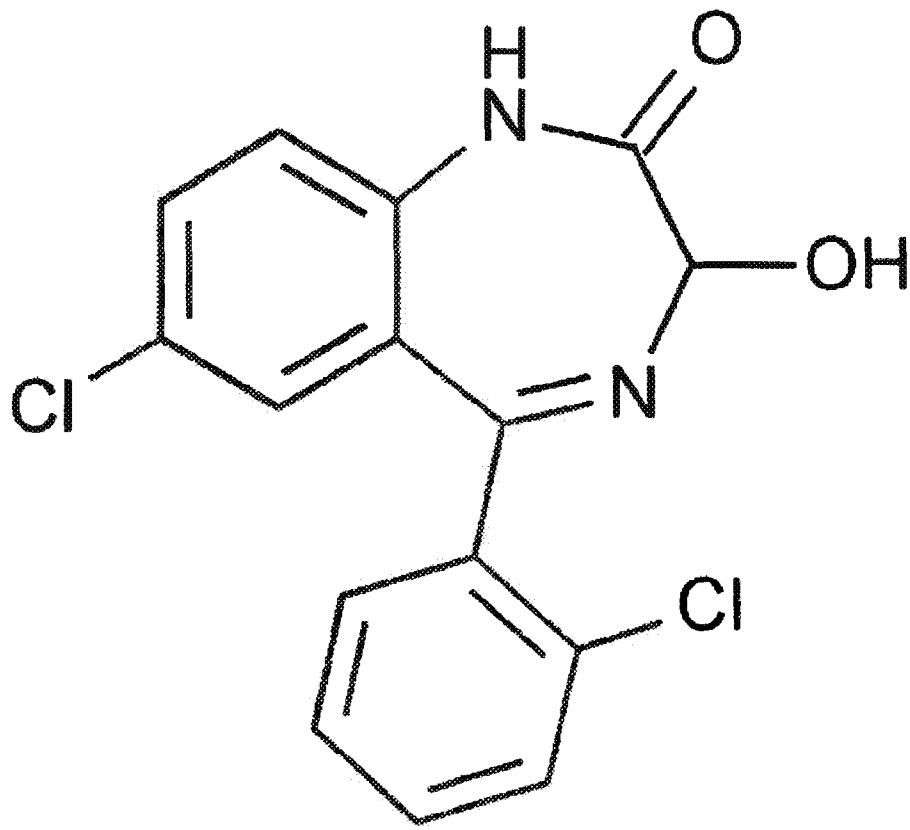
Lorazepam

(lor az' e pam).



USP Image 3D Image

$C_{15}H_{10}Cl_2N_2O_2$ 321.16

2*H*-1,4-Benzodiazepin-2-one, 7-chloro-5-(2-chlorophenyl)-1,3-dihydro-3-hydroxy-, (±)-.
(±)-7-Chloro-5-(*o*-chlorophenyl)-1,3-dihydro-3-hydroxy-2*H*-1,4-benzodiazepin-2-one [846-49-1].

» Lorazepam contains not less than 98.0 percent and not more than 102.0 percent of $C_{15}H_{10}Cl_2N_2O_2$, calculated on the dried basis.

Packaging and storage—Preserve in tight, light-resistant containers.

USP REFERENCE STANDARDS ⟨11⟩—

USP Lorazepam RS

USP Lorazepam Related Compound A RS
7-Chloro-5-(*o*-chlorophenyl)-1,3-dihydro-3-acetoxy-2*H*-1,4-benzodiazepin-2-one.

$C_{17}H_{12}Cl_2N_2O_3$
363.20

USP Lorazepam Related Compound B RS
2-Amino-2′,5-dichlorobenzophenone.
$C_{13}H_9ClNO$
266.13

USP Lorazepam Related Compound C RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxaldehyde.
$C_{15}H_8Cl_2N_2O$
303.15

USP Lorazepam Related Compound D RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxylic acid.
$C_{15}H_8Cl_2N_2O_2$
319.15

USP Lorazepam Related Compound E RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazoline methanol.
$C_{15}H_{10}Cl_2N_2O$
305.16

Identification—

A: *Infrared Absorption* ⟨197K⟩.

B: The retention time of the major peak in the chromatogram of the *Test preparation* corresponds to that in the chromatogram of the *Standard preparation,* as obtained in the *Assay.*

LOSS ON DRYING ⟨731⟩—Dry it in vacuum at 105° for 3 hours: it loses not more than 0.5% of its weight.

RESIDUE ON IGNITION ⟨281⟩: not more than 0.3%.

HEAVY METALS, *Method II* ⟨231⟩: not more than 0.002%.

Related compounds—

Mobile phase and *Diluent*—Prepare as directed in the *Assay.*

Standard solution—Dilute a suitable volume of the *Standard preparation,* prepared as directed in the *Assay,* quantitatively, and stepwise if necessary, to obtain a solution having a known concentration of about 0.032 mg per mL of lorazepam .

Peak identification solution—Dissolve accurately weighed amounts of USP Lorazepam RS, USP Lorazepam Related Compound A RS, USP Lorazepam Related Compound B RS, USP Lorazepam Related Compound C RS, USP Lorazepam Related Compound D RS, and USP Lorazepam Related Compound E RS in *Diluent* to obtain a solution having a final concentration of about 3.2 mg per mL of lorazepam and 0.032 mg per mL each of lorazepam related compound A, lorazepam related compound B, lorazepam related compound C, lorazepam related compound D, and lorazepam related compound E.

Test solution—Dissolve an accurately weighed quantity of Lorazepam in *Diluent,* and dilute quantitatively, and stepwise if necessary, to obtain a solution having a known concentration of about 3.2 mg per mL of lorazepam .

Chromatographic system (see CHROMATOGRAPHY ⟨621⟩)—Proceed as directed in the *Assay.* Chromatograph the *Peak identification solution,* record the peak responses as directed for *Procedure,* and identify the peaks, using the retention times given in *Table 1.*

Table 1

Peak Identification	Approximate Relative Retention Time	Relative Response Factor	Limit (%)
Lorazepam	1.0	1.0	—
Lorazepam related compound D[1]	1.4	1.0	0.15
Lorazepam related compound A[2]	1.7	1.0	0.10
Lorazepam related compound E[3]	1.9	1.3	0.15
Lorazepam related compound C[4]	2.1	1.0	0.30
Lorazepam related compound B[5]	5.5	1.0	0.01
Any individual unspecified impurity	—	1.0	0.10
Total impurities	—	—	0.75

[1] 6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxylic acid.

[2] 7-Chloro-5-(*o*-chlorophenyl)-1,3-dihydro-3-acetoxy-2*H*-1,4-benzodiazepin-2-one.

[3] 6-Chloro-4-(*o*-chlorophenyl)-2-quinazoline methanol.

[4] 6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxaldehyde.

[5] 2-Amino-2′,5-dichlorobenzophenone.

The resolution, *R,* between lorazepam related compound A and lorazepam related

compound E is not less than 1.2. Chromatograph the *Standard solution,* and record the peak responses as directed for *Procedure:* the tailing factor for lorazepam is not more than 2.0 and the relative standard deviation for replicate injections is not more than 5% for lorazepam.

Procedure—Separately inject equal volumes (about 100 µL) of the *Standard solution* and the *Test solution* into the chromatograph, collect the data for at least 50 minutes, and measure the responses for all the peaks. Calculate the percentage of each impurity in the portion of Lorazepam taken by the formula:

$$100(1/F)(C_S/C_U)(r_i/r_S)$$

in which F is the relative response factor for any given impurity found in *Table 1;* C_S and C_U are the concentrations of lorazepam in the *Standard solution* and the *Test solution,* respectively; r_i is the peak response for each impurity in the *Test solution;* and r_S is the peak response for lorazepam obtained from the *Standard solution.* The limit for each related compound is given in *Table 1.*

Assay—

Mobile phase—Prepare a mixture of water, acetonitrile, and glacial acetic acid (50:50:1.2). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Diluent—Prepare a mixture of methanol and water (75:25).

Standard preparation—Dissolve an accurately weighed quantity of USP Lorazepam RS in *Diluent,* and dilute quantitatively, and stepwise if necessary, to obtain a solution having a known concentration of about 0.1 mg of lorazepam per mL.

Assay preparation—Dissolve an accurately weighed quantity of Lorazepam in *Diluent,* and dilute quantitatively, and stepwise if necessary, to obtain a solution having a known concentration of about 0.1 mg per mL of lorazepam.

Chromatographic system (see CHROMATOGRAPHY ⟨621⟩)—The liquid chromatograph is equipped with a sample compartment chiller maintained at 4°, a UV detector set at 230 nm, and a 4.6-mm × 25-cm column that contains 5-µm packing L1. The column temperature is maintained at 5°. The flow rate is about 1.0 mL per minute. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the tailing factor for lorazepam is not more than 2.0, and the relative standard deviation for replicate injections is not more than 2.0% for lorazepam.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, collect the data for at least 50 minutes, and

measure the responses for all the peaks. Calculate the percentage of Lorazepam in the portion of sample taken by the formula:

$$100(C_S / C_U)(r_U / r_S)$$

in which C_S and C_U are the concentrations of lorazepam in the *Standard preparation* and the *Assay preparation,* respectively; and r_U and r_S are the peak responses for lorazepam obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Auxiliary Information— Please check for your question in the FAQs before contacting USP.

Topic/Question	Contact	Expert Committee
Monograph	Hariram Ramanathan, M.S. Associate Scientific Liaison 1-301-816-8313	(SM42010) Monographs - Small Molecules 4
Reference Standards	RS Technical Services 1-301-816-8129 rstech@usp.org	

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Chromatographic Column—

LORAZEPAM

Chromatographic columns text is not derived from, and not part of, USP 34 or NF 29.

Lorazepam Injection

» Lorazepam Injection is a sterile solution of Lorazepam in a suitable medium. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of lorazepam ($C_{15}H_{10}Cl_2N_2O_2$).

Packaging and storage—Preserve in single-dose or multiple-dose containers, preferably of Type I glass, protected from light.

USP REFERENCE STANDARDS ⟨11⟩—

USP Endotoxin RS

USP Lorazepam RS

USP Lorazepam Related Compound B RS
2-Amino-2′,5-dichlorobenzophenone.
$C_{13}H_9ClNO$
266.13

USP Lorazepam Related Compound C RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxaldehyde.
$C_{15}H_8Cl_2N_2O$
303.15

USP Lorazepam Related Compound D RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxylic acid.
$C_{15}H_8Cl_2N_2O_2$
319.15

Identification—

A: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation*, as obtained in the *Assay*.

B: Dissolve USP Lorazepam RS in alcohol to obtain a solution having a concentration of 1 mg per mL. Transfer 10 mL of this solution to a suitable container. Transfer a volume of Injection, equivalent to about 10 mg of lorazepam, to a second container. Separately add 5 mL of hydrochloric acid to each container, heat each solution on a steam bath for 20 minutes, and cool. Transfer the solutions to separators, and add 8 mL of 10 N sodium hydroxide to

each separator. Extract each solution with two 25-mL portions of ether, filtering the ether extracts through cotton plugs into suitable containers. Evaporate both ether extracts to about 2 mL, and add 8 mL of methanol to each. Apply separately 10 µL of the test solution and the Standard solution to a suitable thin-layer chromatographic plate (see *Chromatography* ⟨ 621 ⟩) coated with a 0.25-mm layer of chromatographic silica gel. Allow the spots to dry, and develop the chromatograms in toluene until the solvent front has moved about 15 cm. Remove the plate from the developing chamber, mark the solvent front, and allow the solvent to evaporate. Spray the plate with a freshly prepared 1 in 80 solution of sodium nitrite in 0.5 N hydrochloric acid. Heat the plate at 100° for 5 minutes, allow to cool, and spray with a 1 in 1000 solution of *N*-(1-naphthyl)ethylenediamine dihydrochloride in alcohol: the R_F value of the principal spot obtained from the test solution corresponds to that obtained from the *Standard solution.*

BACTERIAL ENDOTOXINS ⟨ 85 ⟩—It contains not more than 100.0 USP Endotoxin Units per mg of lorazepam .

Related compounds—

A: *Mobile phase, System suitability preparation,* and *Chromatographic system*—Proceed as directed in the *Assay.*

Standard preparation—Prepare a solution in *Mobile phase* having known concentrations of about 3.2 µg each of USP Lorazepam Related Compound C RS and USP Lorazepam Related Compound D RS per mL.

Test preparation—Prepare as directed for *Assay preparation* in the *Assay.*

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Test preparation* into the chromatograph, record the chromatograms, and measure the peak responses of any peaks observed. Do not include as an impurity any peak observed in the chromatogram of the *Test preparation* that has a retention time shorter than that of the lorazepam related compound D peak in the *Standard preparation.* Calculate the percentage of 6-chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxaldehyde (lorazepam related compound C) and the percentage of 6-chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxylic acid (lorazepam related compound D) by the formula:

$$100(C_S / C_U)(r_U / r_S)$$

in which C_S is the concentration, in µg per mL, of the corresponding component in the *Standard preparation;* C_U is the concentration, in µg per mL, of Lorazepam in the *Test preparation;* r_U is the peak response of lorazepam related compound C or lorazepam related compound D in the chromatogram obtained from the *Test preparation;* and r_S is the

peak response of the corresponding component in the *Standard preparation*. Calculate the percentage of any other impurity detected in the chromatogram of the *Test preparation* by the formula:

$$100(r_i / r_T)$$

in which r_i is the peak response of the individual impurity; r_T is the peak response of lorazepam obtained from the *Test preparation*. The total of all impurities detected does not exceed 4.0%.

B: Transfer 5.0 mL of Injection to a suitable separator, and add 50 mL of 0.1 N sodium hydroxide. Extract with three 10-mL portions of chloroform, and collect the chloroform extracts in a second separator. Wash the chloroform extracts with 10 mL of water, and transfer the chloroform extracts to a centrifuge tube. Evaporate the chloroform extracts with the aid of a current of air to dryness, and dissolve the residue in acetone to obtain a *Test preparation* having a concentration of 10 mg per mL. Dissolve USP Lorazepam Related Compound B RS in acetone to obtain a *Standard preparation* having a known concentration of 0.1 mg per mL. Apply separately 50 µL of the *Test preparation* and 5 µL of the *Standard preparation* to a suitable thin-layer chromatographic plate (see *Chromatography* ⟨621⟩) coated with a 0.25-mm layer of chromatographic silica gel mixture. Allow the spots to dry, and develop the chromatograms in a solvent system consisting of a mixture of chloroform, *n*-heptane, and alcohol (10:10:1) until the solvent front has moved not less than 10 cm from the origin. Remove the plate from the developing chamber, mark the solvent front, and allow the solvent to evaporate. Lightly spray the plate with 2 N sulfuric acid, dry at 105° for 15 minutes, and spray successively with sodium nitrite solution (1 in 1000), ammonium sulfamate solution (1 in 200), and *N*-(1-naphthyl)ethylenediamine dihydrochloride solution (1 in 1000), drying the plate with a current of air after each spraying. Observe the plate under visible light: the spot produced by the *Test preparation* is not greater in size or intensity than the principal spot produced at the corresponding R_F value by the *Standard preparation*, corresponding to not more than 0.1% of 2-amino-2′,5-dichlorobenzophenone (lorazepam related compound B).

Other requirements—It meets the requirements under *Injections* ⟨1⟩.

Assay—

Mobile phase—Prepare a mixture of methanol and 0.05 M monobasic ammonium phosphate (50:50). Adjust with ammonium hydroxide to a pH of 6.5, filter, and degas. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve an accurately weighed quantity of USP Lorazepam RS in methanol to obtain a solution having a known concentration of about 1.0 mg per mL. Transfer 4.0 mL of this solution to a 25-mL volumetric flask, dilute with *Mobile phase* to volume, and

mix to obtain a solution having a known concentration of about 0.16 mg per mL.

Assay preparation—Transfer an accurately measured volume of Injection, equivalent to about 4 mg of lorazepam, to a 25-mL volumetric flask, dilute with *Mobile phase* to volume, and mix.

System suitability preparation—Prepare a solution of Lorazepam in *Mobile phase* containing about 0.04 mg of lorazepam per mL and about 0.032 mg each of lorazepam related compound C and lorazepam related compound D per mL.

Chromatographic system (see Chromatography ⟨621⟩)—The liquid chromatograph is equipped with a 240-nm detector and 4.6-mm × 10- to 15-cm column that contains packing L1. The flow rate is about 2 mL per minute. Chromatograph replicate injections of the *Standard preparation*, and record the peak responses as directed for *Procedure:* the relative standard deviation is not more than 2.0%. Chromatograph the *System suitability preparation*, and record the peak responses as directed for *Procedure:* the resolution, *R,* between any of the major peaks is not less than 1.2; and the relative retention times are about 0.7 for 6-chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxylic acid (lorazepam related compound D), 1.0 for lorazepam, and 2.7 for 6-chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxaldehyde (lorazepam related compound C).

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the responses for the major peaks. Calculate the quantity, in mg, of lorazepam ($C_{15}H_{10}Cl_2N_2O_2$) in each mL of the Injection taken by the formula:

$$25(C / V)(r_U / r_S)$$

in which C is the concentration, in mg per mL, of USP Lorazepam RS in the *Standard preparation; V* is the volume, in mL, of Injection taken; and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation*, respectively.

Auxiliary Information— Please check for your question in the FAQs before contacting USP.

Topic/Question	Contact	Expert Committee
Monograph	Hariram Ramanathan, M.S. Associate Scientific Liaison 1-301-816-8313	(SM42010) Monographs - Small Molecules 4
Reference Standards	RS Technical Services 1-301-816-8129 rstech@usp.org	
⟨85⟩	Radhakrishna S Tirumalai, Ph.D. Principal Scientific Liaison	(GCM2010) General Chapters - Microbiology

	1-301-816-8339	

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Chromatographic Column—

LORAZEPAM INJECTION

Chromatographic columns text is not derived from, and not part of, USP 34 or NF 29.

Lorazepam Oral Concentrate

» Lorazepam Oral Concentrate contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of lorazepam ($C_{15}H_{10}Cl_2N_2O_2$).

Packaging and storage—Preserve in well-closed, light-resistant containers.

USP REFERENCE STANDARDS ⟨ 11 ⟩—

USP Lorazepam RS

USP Lorazepam Related Compound B RS
2-Amino-2′,5-dichlorobenzophenone.
$C_{13}H_9ClNO$
266.13

USP Lorazepam Related Compound C RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxaldehyde.
$C_{15}H_8Cl_2N_2O$
303.15

USP Lorazepam Related Compound D RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazolinecarboxylic acid.
$C_{15}H_8Cl_2N_2O_2$
319.15

USP Lorazepam Related Compound E RS
6-Chloro-4-(*o*-chlorophenyl)-2-quinazoline methanol.
$C_{15}H_{10}Cl_2N_2O$
305.16

Identification—The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that of the *Standard preparation* as obtained in the *Assay*.

Related compounds—

Mobile phase—Prepare a mixture of methanol and 0.05 M monobasic ammonium phosphate (64:36). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨ 621 ⟩).

System suitability preparation—Proceed as directed in the *Assay* under *Lorazepam Injection*.

Standard solution—Prepare a solution in *Mobile phase* having known concentrations of about 3.2 µg each of USP Lorazepam Related Compound C RS and USP Lorazepam Related Compound D RS, and 0.16 µg of USP Lorazepam Related Compound B RS per mL.

Test solution—Transfer an accurately measured volume of Oral Concentrate, equivalent to about 4 mg of lorazepam , to a 25-mL volumetric flask, dilute with *Mobile phase* to volume, and mix to obtain a solution having a known concentration of about 0.16 mg per mL.

Chromatographic system—Proceed as directed in the *Assay* under *Lorazepam Injection*, except that the flow rate is 0.7 mL per minute.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard solution* and the *Test solution* into the chromatograph, record the chromatograms, and measure the responses of any peaks other than the lorazepam peak. Do not include as an impurity any peak observed in the chromatogram of the *Test solution* that has a retention time shorter than that of the lorazepam related compound D peak in the *Standard solution*. Calculate the percentages of lorazepam related compound B, lorazepam related compound C, and lorazepam related compound D taken by the formula:

$$100(C_S / C_U)(r_U / r_S)$$

in which C_S is the concentration, in µg per mL, of the corresponding component in the *Standard solution*; C_U is the concentration, in µg per mL, of lorazepam in the *Test solution*; r_U is the peak response of lorazepam related compound B, lorazepam related compound C, or lorazepam related compound D in the chromatogram obtained from the *Test solution*; and r_S is the peak response of the corresponding component in the *Standard solution*. Not more than 0.1% of lorazepam related compound B is found; and not more than 4.0% for the sum of lorazepam related compound C and lorazepam related compound D is found.

Assay—

Mobile phase—Prepare a mixture of water, acetonitrile, and glacial acetic acid (55:45:0.2). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

System suitability preparation—Dissolve 10 mg each of Lorazepam and USP Lorazepam Related Compound E RS in 100 mL of methanol.

Standard preparation—Dissolve an accurately weighed quantity of USP Lorazepam RS in methanol, and dilute quantitatively, and stepwise if necessary, with methanol to obtain a solution having a known concentration of about 0.05 mg per mL.

Assay preparation—Transfer an accurately measured volume of Oral Concentrate, equivalent to 5 mg of lorazepam , to a 100-mL volumetric flask, and dilute with methanol to

volume to obtain a solution containing about 0.05 mg of lorazepam per mL.

Chromatographic system (see **CHROMATOGRAPHY** ⟨ 621 ⟩)—The liquid chromatograph is equipped with a 254-nm detector and a 4-mm × 30-cm column that contains packing L1. The flow rate is about 2 mL per minute. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the relative standard deviation is not more than 2.0%. Chromatograph the *System suitability preparation,* and record the peak responses as directed for *Procedure:* the relative retention times are about 0.6 for lorazepam and 1.0 for lorazepam related compound E; and the resolution, *R,* between lorazepam and lorazepam related compound E is not less than 2.0.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the responses for the major peaks. Calculate the quantity, in mg, of lorazepam ($C_{15}H_{10}Cl_2N_2O_2$) in the portion of Oral Concentrate taken by the formula:

$$100C(r_U / r_S)$$

in which C is the concentration, in mg per mL, of USP Lorazepam RS in the *Standard preparation;* and r_U and r_S are the lorazepam peak responses obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Auxiliary Information— Please check for your question in the FAQs before contacting USP.

Topic/Question	Contact	Expert Committee
Monograph	Hariram Ramanathan, M.S. Associate Scientific Liaison 1-301-816-8313	(SM42010) Monographs - Small Molecules 4
Reference Standards	RS Technical Services 1-301-816-8129 rstech@usp.org	

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Chromatographic Column—

LORAZEPAM ORAL CONCENTRATE

Chromatographic columns text is not derived from, and not part of, USP 34 or NF 29.